12-10-2020

Isolation of Halotolerant Bacteria from the Rhizosphere of Ceanothus velutinus May Lead to Contributions in Plant Health in Saline Conditions

Jacob Davis
Utah State University, jathayd@gmail.com

Isolation of halotolerant bacteria from the rhizosphere of *Ceanothus velutinus* may lead to contributions in plant health in saline conditions.

Jacob Davis[2], Jyothsna Genesh[1], Amita Kaundal[1*]

Department of Plant, Soil, and Climate[1], CAAS, Department of Biology[2], CS
Utah State University, Logan, Utah.

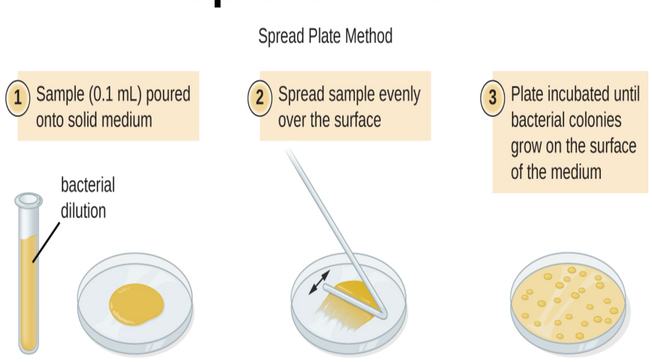
INTRODUCTION

The recent rise in the average global temperature has been a driving force over the past few years for rising soil salinity. This condition presents a hostile environment to many plant species that have not previously been exposed to these conditions. The rhizosphere, a layer of soil attached to the roots of a plant, contains microorganisms that may contribute to the plants' abiotic and biotic stress resistance. These microorganisms are known as Plant Growth Promoting Rhizobacteria (PGPR). These can play a significant role in contributing to plant stress resistance. Some native plants have shown a strong ability to resist harsh or acclimate abiotic and biotic stressors such as drought, cold temperatures, heavy metal contaminations, and more. In this study, we have selected a native resilient plant indigenous to the InterMountain West region of North America, known as *Ceanothus velutinus* (Snowbrush). We aim to try to isolate halotolerant bacteria from the rhizosphere of Snowbrush. Samples were collected from the rhizosphere of Snowbrush plants from the Tony Grove region of Logan Utah.

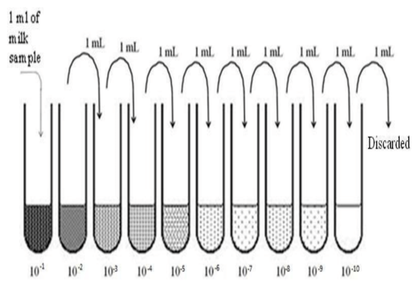
METHODOLOGY

The root samples of the Snowbrush plant from three different locations in Tony Grove, Utah were collected. Rhizosphere soil separated from the roots. The soil samples were diluted to a 10:95 ratio in water. These dilutions further serially diluted five times in a 1:10 ratio. y diluted five times in a 1:10 ratio.

Spread Plate



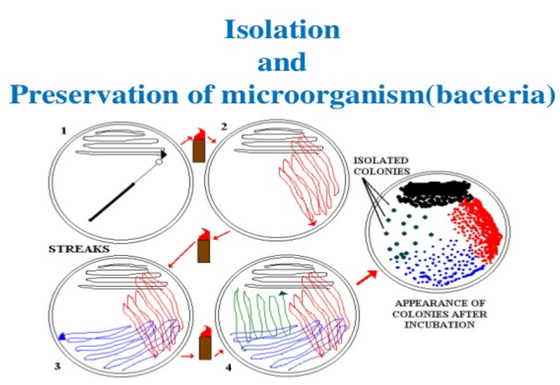
Serial Dilution



The dilutions of 10^-3, 10^-4, 10^-5 were then plated onto nutrient agar media with six different concentrations of sodium chloride (2%, 4%, 6%, 8%, 10% W/V) with 0% as a control.

Once colonies have grown on the media, each bacterial colony is streaked and purified. The isolated colonies will be identified using 16s rRNA sequencing then followed by BLAST (Basic local alignment search tool) search.

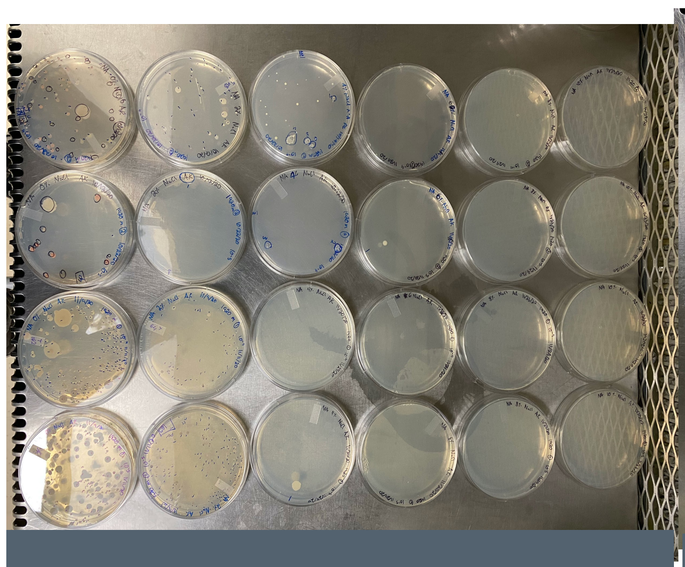
Streak Plate



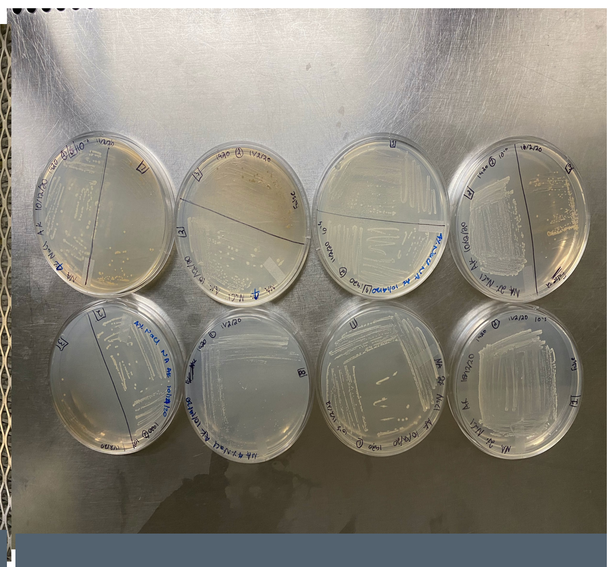
RESULTS

Sample	0% NaCl 10^-4	0% NaCl 10^-5	2% NaCl 10^-4	2% NaCl 10^-5	4% NaCl 10^-4	4% NaCl 10^-5	6% NaCl 10^-4	6% NaCl 10^-5	8% NaCl 10^-4	8% NaCl 10^-5	10% NaCl 10^-4	10% NaCl 10^-5
1920 (1)	162	185	64	160	0	1	0	0	0	0	0	0
1920 (2)	73	12	48	1	31	2	0	1	0	0	0	0

Table 1: Total number of colonies observed in each of sample and concentration plated.



Bacterial growth on all the plates.



Purified colonies from the 2 % and 4% salt concentrations.

A rhizosphere soil sample from two plants of one location 1920m was grown on nutrient agar media with 6 different concentrations of sodium chloride.
All 0% concentrations were used as a control and in each of those plates there were dozens to hundreds of colonies isolated depending on the sample. At 2% NaCl concentration there was still a very high amount of colonies being able to grow in the media. For example 1920 (2) 10^-4 in 2% had 48 colonies, and 1920 (1) 10^-5 in 2% had 160 colonies. As concentrations increased the number of colonies able to proliferate decreased. By 4% NaCl most samples were down to or near only a single colony being manifested. With one exception being 1920m (2) 10^-4 on 4% having 31 colonies. Only one sample managed to grow a single colony in 6%. That being 1920m (2) 10^-5. No colonies were observed at or past 8% NaCl concentration. Each unique colony type from each media and dilution were streaked on the same media and concentration in order to isolate a single species of bacteria to a plate. Sequencing of these bacterial species for the 16SrRNA region is under process.

DISCUSSION

The purpose of this study is to isolate halotolerant bacteria from native plants that survive in harsh conditions. After isolation, purification, and identification, these microbes will be tested on the model plant *Arabidopsis thaliana* with various salt treatments to see their role in the plant's growth and development under salt stress.

Utah State University
DEPARTMENT OF PLANTS, SOILS, & CLIMATE

Jacob Athay Davis
360-984-4731
jathayd@gmail.com
USU College of Science
Department of Biology